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18. This work was supported by the Office of Naval Research, the National Science Foundation, and the Robert A. Welch Foundation. Discussions with J. L. Martins are gratefully acknowledged, as is the ongoing cheerful support of Park Scientific Instruments.

28 May 1991; accepted 19 June 1991

Density-Dependent Natural Selection and Trade-Offs in Life History Traits

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Theories of density-dependent natural selection state that at extreme population densities evolution produces alternative life histories due to trade-offs. The trade-offs are presumed to arise because those genotypes with highest fitness at high population densities will not also have high fitness at low density and vice-versa. These predictions were tested by taking samples from six populations of *Drosophila melanogaster* kept at low population densities (*r*-populations) for nearly 200 generations and placing them in crowded cultures (*K*-populations). After 25 generations in the crowded cultures, the derived *K*-populations showed growth rate and productivity that at high densities were elevated relative to the controls, but at low density were depressed.

ONE OF THE FIRST SUCCESSFUL combinations of theory from ecology and evolution was the theory of density-dependent natural selection, often called *r*- and *K*-selection, where *r* and *K* refer to low- and high-density conditions, respectively (*1*). The initial models combined theoretical models of population growth dynamics with single-locus population genetic models in order to describe evolution in environments that differ with respect to population density (*2*); more complex elaborations of these early models were advanced later (*3*). These theories and models have assumed a central role in the theory of evolutionary ecology (*4*). A crucial assumption of these theories has been that genotypes selected for sustained reproduction and survival at high population densities are not likely to do as well at low densities; likewise organisms capable of rapid reproduction under low levels of crowding may not reproduce as well under crowded conditions. It has been, however, difficult to demonstrate empirically the postulated trade-offs (*5*).

We have previously shown (*6*) that three populations of *Drosophila melanogaster* kept at low density (*r*-populations) had, after eight generations, higher rates of population growth when tested at low densities than three populations kept at high density (*K*-populations), whereas the opposite was the case for growth rates tested at high population densities. The interpretation and significance of these results were complicated by three issues. (i) Given that both the low- and high-density environments were novel for these populations, it remains possible that the differences observed between the *r*- and *K*-populations were not due to changes in both populations but rather that only one population had evolved whereas the other retained the attributes of the founder population. (ii) The differences observed between *r*- and *K*-populations with respect to growth rates and total productivity at low density were only marginally significant. (iii) No additional tests have been carried out to verify these results; indeed one study of mosquitoes from natural populations did not show any trade-off in population growth rates (*7*).

We describe an experiment designed to overcome these problems, the results of which confirm that trade-offs do occur in the evolution by density-dependent natural selection. We test two types of high density populations (*rK*- and *r*×*rK*-populations), both derived from *r*-populations now transferred to the *K*-environment; the controls are two types of low density populations (*r*- and *r*×*r*-populations) (Fig. 1). The *r*×*r*-populations were created to introduce genetic variation into replicate sets of low density populations. The *rK*-populations were created from samples of each *r*-population and had been maintained at high densities for 25 generations prior to this experiment. In a similar fashion the *r*×*rK*-populations are samples from the *r*×*r*-populations that have been kept at high densities.

Each of the four types of populations consisted of three replicates. During the course of this experiment the *r*- and *r*×*r*-populations are not expected to change significantly, given that they had been previously kept for nearly 200 generations at the same low density as now. However, the *rK*- and *r*×*rK*-populations have been transferred from the low density *r*-environment to the high density *K*-environment. Hence, differences that arise between them and the *r* and *r*×*r* controls may be attributed to adaptation to the new high-density environments.

After the three *rK*- and three *r*×*rK*-populations had undergone 25 generations of natural selection in their new environments, we measured rates of population growth and net productivity in each of these six

Table 1. The mean values (and standard errors) for productivity and growth rate for each population and each density.

Population	Productivity (no. of adults) by density			Growth rate (no. of offspring per adult per week) by density		
	10	750	1000	10	750	1000
r_1	872 (52)	739 (24)	817 (17)	5.58 (0.20)	1.53 (0.017)	1.40 (0.021)
r_2	917 (16)	800 (38)	808 (20)	6.01 (0.16)	1.55 (0.03)	1.43 (0.0075)
r_3	788 (21)	770 (47)	797 (45)	5.05 (0.19)	1.47 (0.033)	1.37 (0.019)
rK_1	834 (26)	784 (24)	858 (67)	5.69 (0.10)	1.51 (0.015)	1.43 (0.029)
rK_2	799 (22)	856 (9)	935 (23)	5.19 (0.17)	1.52 (0.009)	1.45 (0.012)
rK_3	725 (41)	852 (14)	948 (33)	5.09 (0.15)	1.52 (0.0031)	1.43 (0.0092)
$r \times r_1$	736 (20)	689 (28)	823 (20)	5.54 (0.12)	1.43 (0.016)	1.37 (0.0044)
$r \times r_2$	772 (28)	745 (24)	846 (52)	5.16 (0.095)	1.42 (0.0088)	1.37 (0.022)
$r \times r_3$	774 (16)	743 (10)	822 (61)	5.38 (0.12)	1.41 (0.0048)	1.35 (0.026)
$r \times rK_1$	680 (19)	773 (24)	862 (18)	5.13 (0.037)	1.45 (0.0062)	1.40 (0.011)
$r \times rK_2$	699 (32)	804 (18)	952 (47)	5.11 (0.10)	1.44 (0.012)	1.41 (0.026)
$r \times rK_3$	667 (32)	799 (38)	944 (24)	4.98 (0.12)	1.45 (0.017)	1.39 (0.0031)

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populations and the six controls, at three different densities (10, 750, and 1000 adults) following the methods described in (*6*). We determined the rates of population growth (*6*) and the net productivity (*8*), two different measures of population fitness that can be estimated from the same experimental data (Table 1 and Figs. 2 and 3). Rate of population growth is the measure of fitness most closely tied to the theories of density-dependent natural selection, but it cannot be estimated as accurately as can net productivity. Trade-offs should produce a significant interaction between density and population in an analysis of variance (ANOVA). With the sample sizes used in this study the power of the typical ANOVA is quite low, so we have also tested the differences in growth rate and productivity by the Mann-Whitney *U* test.

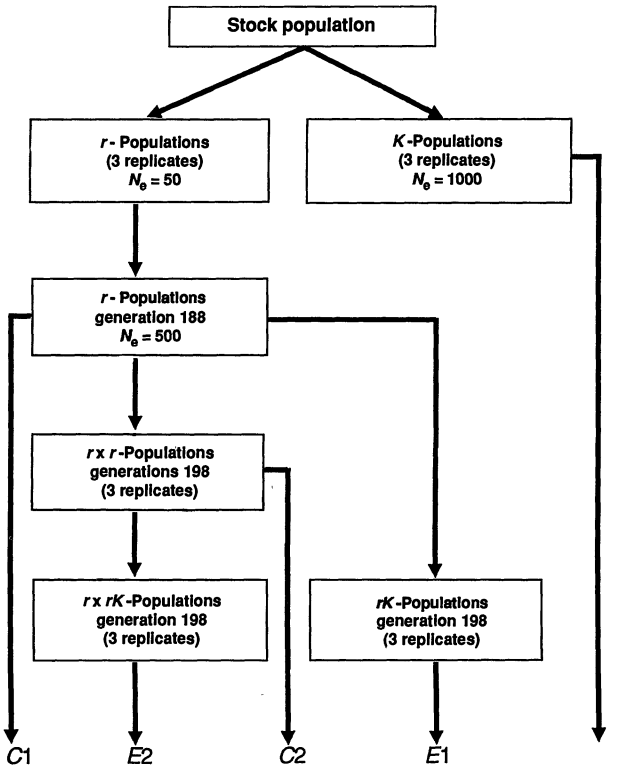

Fig. 1. Derivation of the experimental stocks used in this study. In November 1978 the *r*- and *K*-populations were initiated from random samples of a genetically diverse population of *D. melanogaster* (derived from more than 1000 gravid females collected in Alameda County, California). The three replicate *r*-populations (r_1, r_2, r_3) and the three replicate *K*-populations (K_1, K_2, K_3) have been continuously maintained independently of each other. The *K*-populations are maintained at high population densities by a serial transfer system, whereas the *r*-populations are maintained at low population densities on a discrete life cycle (*6*). During the first 188 generations the effective population size (N_e) of the *r*-populations was about 50. After this time their effective size was increased to 500, to forestall any further effects of random genetic drift, while keeping the levels of adult and larval crowding identical to those in the previous 188 generations. At generation 198 the three *r*-populations were crossed in all possible pairwise combinations so as to increase the genetic diversity in any one by combining all three. Approximately equal numbers of progeny were collected from the six different crosses to initiate each of three new populations, called *r*×*r*. Large egg samples were taken from each of the three *r*-populations and used to start three *rK*-populations that are maintained at high densities in the same way as the *K*-populations: rK_1 is the population derived from r_1, rK_2 is derived from r_2, and rK_3 from r_3. Large egg samples from the *r*×*r*-populations were taken to initiate three *r*×*rK*-populations. The experiments reported here were done in generation 223 after the *r*- and *K*-populations were initiated. Thus, the *rK*- and *r*×*rK*-populations had completed 25 generations in the *K*-environment. In this experiment control C1 is compared to experimental population *E*1 and *C*2 to *E*2.

The differences between the *r*×*rK* and the *r*×*r*-populations in net productivity are statistically significant at all densities (Fig. 2). The expected trade-offs are evident. At low density (ten adults per culture) the net production of progeny is greater in the *r*×*r*-populations than in the *r*×*rK*-populations, whereas the reverse occurs at the two high densities. Similar trade-offs in the net productivity are apparent between the *r*- and *rK*-populations, but because of high within-treatment variability only the difference at the highest density is statistically significant.

The expected trade-offs also occur with respect to population growth rate (Fig. 3). The differences between the *r*×*rK*- and *r*×*r*-populations are each statistically significant by at least one test. The *rK*-populations show a significantly higher average growth rate than their *r* controls at the highest density, but no significant differences occur at the two other densities (Fig. 3).

The growth rate data of the *r*- and *rK*-

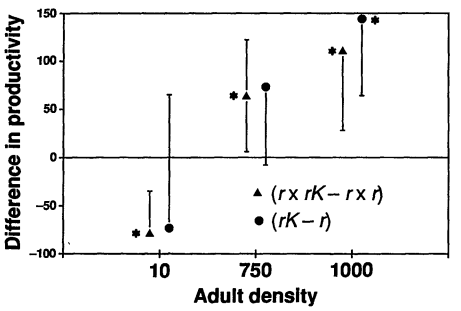

Fig. 2. Experiments to measure net productivity. These were conducted as follows. Adults that have passed through two generations in common conditions were placed in half-pint cultures with standard medium at one of three densities (10,750, or 1,000). After 1 week of egg laying the adults are removed and the number of survivors counted. The progeny emerging from each bottle were counted at weekly intervals for the next 3 weeks. Net productivity is the difference between the sum of the survivors plus progeny and the initial density of adults. Rates of population growth depend on both the number of surviving adults and progeny produced. Hence, both these quantities are used in these estimates of net productivity although other investigators have concentrated on just progeny production (*8*). For each population, six replicate experiments were conducted at density 10, and three at each of the two higher densities. The graph shows the difference in net productivity between the mean of the three *K*-populations (either *rK* or *r*×*rK*) and the three controls (*r* or *r*×*r*). Only the plus or minus portion of the 95% confidence interval is shown when one of the bars would extend beyond the graph. The null hypothesis is that the *K*-populations do not have lower productivity than the controls in the low-density tests and higher in the high-density tests. The tests reject this null hypothesis, thereby supporting our working hypothesis that trade-offs in life history evolve by natural selection in response to population density. The asterisk indicates that the difference is statistically significant according to the Mann-Whitney *U* test ($P = 0.05$). The ANOVA gave significant population-by-density interactions for *r* versus *rK* ($P = 0.002$), and for *r*×*r* versus *r*×*rK* ($P < 0.001$). The difference in the ordinate is in numbers of individuals.

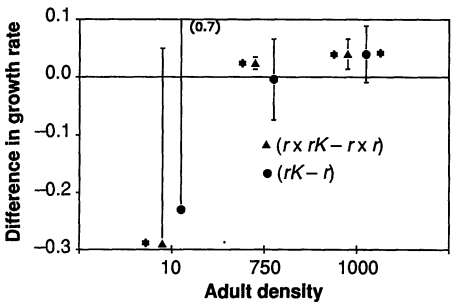

Fig. 3. Growth rate is measured in units of number of offspring per adult per week. The differences illustrated are from the experiments described in the legend to Fig. 2. Growth rate takes into account the time when progeny are produced (*6*) in addition to just the number of survivors and offspring (which are the components of net productivity). Thus, progeny that emerge during the second week of the experiment will have a greater impact on the growth rate than progeny that emerge during the fourth week. For the computation of net productivity the progeny from the second and fourth weeks have equal effects. The symbols indicate statistical significance as described for Fig. 2. The ANOVA, on log-transformed data, indicates no significant population by density interaction for *r* versus *rK* but a significant interaction for *r*×*r* versus *r*×*rK* ($P = 0.003$). These experiments, like the tests described in Fig. 2, corroborate the evolution of life history trade-offs in response to population density. The 95% confidence interval is shown for differences in growth rate.

populations show significant within-population heterogeneity. This may be because the three *r*-populations became genetically differentiated from each other due to random fixation of alleles during the 188 generations preceding the present experiment. The *rK*-populations derived from them would be similarly genetically heterogeneous. This between-population variation would affect the measured traits. We can deal with this by comparing each replicate *rK*-population to the *r*-population from which it was derived. These comparisons yield no significant difference in growth rate at the lowest density between r_1 and rK_1 or r_3 and rK_3 but the difference between rK_2 and r_2 (−0.82) is statistically significant.

Our previous work has shown that certain larval characters, such as larval feeding rates and pupation site choice have become differentiated between the *r*- and *K*-populations (*9–11*). These behavioral traits contribute to adaptation at high population density: in food-limited environments increased pupation height increases viability (*10*) and increased larval feeding rate increases larval competitive ability (*11*). These characters have also become differentiated between the *r*- and *rK*-populations (*12*), confirming that the *rK*-populations are adapting to these crowded environments.

The results of the experiments reported here confirm earlier observations of fitness trade-offs arising from density-dependent natural selection (*6*). These experiments also show that these trade-offs can arise simply from the process of *Drosophila* adapting to crowded population conditions. The *r*-populations has spent nearly 200 generations in the low-density conditions. The possibility that during the course of the current experiment (generation 201 to 225 in the *r*-populations) there were further major phenotypic changes in the *r*- or *r*×*r*-populations is minimal. Consequently, the changes now observed between the *r*- and *rK*-populations and between the *r*×*r*- and *r*×*rK*-populations may be safely attributed to changes in the high-density populations as they adapt for 25 generations to these novel environments.

As pointed out above, the primary characters responsible for the differences in population growth rate seem to be larval attributes. These larval differences have presumably developed in response to the high larval densities. Although there are differences in the timing of adult reproduction in the *r*- and *K*-populations (*13*), the likelihood that these time differences influence the evolution of the larval characters is minimal because they should have no effect on larval densities.

Much of the current theory in life history evolution assumes that trade-offs in fitness components are important determinants that constrain the direction of evolution (*14*). For instance, in *Drosophila* populations that experience a wide range of population densities through time, neither the *r* phenotype or the *K* phenotype described here would be most fit at all times. There are few well-documented examples of such trade-offs during evolution. Rose (*15*) has shown that natural selection for increased longevity in *D. melanogaster* has been accompanied by a decline in early fecundity of females. The possibility exists that natural selection may act further to offset the maladaptive features of trade-offs (*16*). Our populations of *Drosophila* represent one of the few instances in which trade-offs have been repeatedly demonstrated to affect the outcome of natural selection. Understanding the genetic and physiological underpinnings of these trade-offs becomes an important goal for future research in evolutionary ecology.

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17. We thank M. Rose and three referees for useful comments on the manuscript and R. McCleary for statistical advice. Supported in part by a grant from the Department of Energy and by NIH grant BRSG S07 RR07008.

15 January 1991; accepted 22 May 1991

Conversion of Ectoderm to Mesoderm by Cytoplasmic Extrusion in Leech Embryos

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The role of cytoplasmic domains in the determination of the fates of ectodermal and mesodermal cells has been investigated in leech embryos. When yolk-deficient cytoplasm (teloplasm) was extruded from the animal pole of the zygote, the ectodermal precursor blastomere was converted to a mesodermal fate. This change of fate can be prevented by replacement of the extruded animal teloplasm with teloplasm from the vegetal pole. The fate of the mesodermal precursor blastomere was unaffected by teloplasm extrusion or rearrangement. These results demonstrate that ectodermal and mesodermal determination of fate involves a binary decision dependent on the position of teloplasm along the animal-vegetal axis.

THE SPECIFICATION OF ECTODERMAL and mesodermal fate occurs early in the development of most animals and is achieved by a variety of mechanisms, including the segregation of localized determinants during cleavage (*1*). Like many invertebrates, glossiphoniid leeches develop in a highly determinate manner, meaning that early cell divisions are stereotyped and give rise to unique cells with predictable fates (*2–4*). The separation of segmental ectodermal and mesodermal lineages occurs with the cleavage of a single cell, when blastomere D′ gives rise to cells DNOPQ and DM (*5*) at the fourth cleavage (Fig. 1A). DNOPQ, which is situated more toward the animal pole than DM, subsequently produces four bilateral pairs of ectodermal stem cells (the N, O/P, O/P, and Q ectoteloblasts) and 13 micromeres, whereas DM, which is situated more toward the vegetal pole than DNOPQ, gives rise to one bilateral pair of mesodermal stem cells (mesoteloblasts) and two micromeres (*6*). Each teloblast undergoes a series of highly unequal divisions to generate a chain (bandlet) of smaller blast cells. Subsequently, the left and right bandlets coalesce along the ventral midline into a sheet of cells called the germinal plate

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